

## Identification and Pathogenicity of *Botryosphaeriaceae* Species Associated with Canker Diseases in Redbuds

### Abstract:

Redbuds (*Cercis* sp.), valued for their vibrant flowers and heart-shaped foliage, are economically important trees in Tennessee nurseries. However, their production is challenged by wood canker and dieback diseases caused by fungal pathogens in the *Botryosphaeriaceae* family. There is limited taxonomic information about the fungal species that cause canker diseases in redbuds. This study aimed to identify and characterize the major fungal canker pathogens affecting redbuds in Tennessee. From 2022 to 2024, the plant pathology laboratory in McMinnville, Tennessee, received over 100 redbud plants exhibiting canker symptoms from various commercial nurseries in Tennessee. The symptoms included dark sunken lesions, shoot dieback, thickened callus, and V-shaped discoloration in woody tissues. Samples were processed for the morphological, microscopic, and molecular characterization of the causal agents. Colony morphology, pycnidia, and conidial characteristics were observed for morphological and molecular identification, while phylogenetic analysis was performed by PCR amplification of the *ITS*,  *$\beta$ -tubulin*, and *TEF1* genes. Botryosphaeriaceae accounted for 40% of all isolates. Four major Botryosphaeriaceae species, *Botryosphaeria dothidea*, *Diplodia seriata*, *Lasiodiplodia theobromae*, and *Neofusicoccum parvum* were identified. Among these, *B. dothidea* was the most frequently isolated species. Pathogenicity tests were conducted twice in a greenhouse between June and November of 2023 and 2024, following a completely randomized design. There were six replicates per treatment, and healthy 1-year-old eastern redbud plants were artificially inoculated using mycelial plugs. The pathogenicity test showed the pathogenic nature of all four tested isolates with varying degrees of virulence. Lesion lengths measured six months post-inoculation showed that *L. theobromae* was the most aggressive species, producing the largest mean lesion length (5.62 cm) with a pathogen recovery percentage of 98%, followed by *N. parvum* (3.47 cm, 88%), *B. dothidea* (2.75 cm, 86%) and *D. seriata* (1.6 cm, 66%). This study shows the prevalence, diversity, and virulence of Botryosphaeriaceae species affecting redbuds in Tennessee nurseries. These findings will enhance our understanding of these pathogens and support the development of effective strategies to manage canker diseases caused by *Botryosphaeriaceae* in redbud nurseries.